Legionnaires' Disease in South Africa, 2012–2014

Nicole Wolter, Maimuna Carrim, Cheryl Cohen, Stefano Tempia, Sibongile Walaza, Philip Sahr, Linda de Gouveia, Florette Treurnicht, Orienka Hellferscee, Adam L. Cohen, Alvaro J. Benitez, Halima Dawood, Ebrahim Variava, Jonas M. Winchell, Anne von Gottberg

During June 2012–September 2014, we tested patients with severe respiratory illness for *Legionella* spp. infection and conducted a retrospective epidemiologic investigation. Of 1,805 patients tested, *Legionella* was detected in samples of 21 (1.2%); most were adults who had HIV or tuberculosis infections and were inappropriately treated for *Legionella*.

Data are limited regarding prevalence of *Legionella* spp. bacteria that cause community-acquired pneumonia (CAP) in Africa (*1*), despite the high prevalence of HIV-infected adults in many African countries, including South Africa (*2*). Legionellosis is a notifiable disease in South Africa but is rarely reported. We sought to determine the prevalence of *Legionella* spp. infections in South Africa and describe epidemiologic characteristics of patients with Legionnaires' disease (LD).

The Study

During June 2012–September 2014, we conducted a prospective, hospital-based, observational study as part of the severe respiratory illness (SRI) surveillance at 2 sites in South Africa: Klerksdorp-Tshepong Hospital Complex, Klerksdorp, North West Province; and Edendale Hospital, Pietermaritzburg, KwaZulu-Natal Province. A patient with SRI was defined as a person hospitalized with lower respiratory tract infection of any duration. We used a standardized questionnaire to collect demographic and clinical information. Nasopharyngeal specimens and induced sputum samples were tested for *Legionella* spp. infections by using a real-time PCR assay, as previously described (*3*). Specimens that were *Legionella* positive were also tested by real-time PCR assays to identify *L. pneumophila* and *L. longbeachae*. In addition, patients' specimens were tested for other respiratory pathogens and for HIV. Of the 22 *Legionella*-positive patients, we could trace 17 with whom we conducted a retrospective epidemiologic investigation, which included interviews (detailed study methods in the online Technical Appendix, http://wwwnc.cdc.gov/EID/article/22/1/15-0972-Techapp.pdf).

During June 2012–September 2014, a total of 4,525 SRI patients were enrolled; induced sputum specimens, the recommended specimen type for *Legionella* spp. detection, were collected from 1,805 (40%). Of 1,803 patients with sputum specimens for which data were available, 885 (49%) were male, and 324 (18%) were children <5 years of age. HIV prevalence was 64% (1,025 of 1,594 patients with sputum specimens and known HIV status), and prevalence of active tuberculosis (TB) infection was 24% (421 of 1,758 patients with sputum specimens and known TB status). Of 1,720 patients with sputum specimens and known survival status, 142 (8%) patients died.

Among the 1,805 patients with sputum samples, 21 (1.2%, 95% CI 0.7%–1.7%) tested positive for *Legionella* spp. by real-time PCR. For 1 patient (designated E1 in the online Technical Appendix Table) from whom sputum could not be collected, *Legionella* spp. infection was detected in the nasopharyngeal specimen, so 22 patients with *Legionella* spp. infections were detected in total. Among the 21 patients whose sputum tested positive for *Legionella* spp. infections, median age was 40 years (range 19–59 years; Figure 1), and 11 (52%) were males.

A cluster of case-patients (15/21 [71%]) was observed during July–December 2012 (Figure 2), including all 6 from Edendale Hospital and 10 (10/16, 63%) from Klerksdorp-Tshepong Hospital Complex. These sites are geographically distant (≈600 km) from one another, so the respective clusters or outbreaks are unlikely to be related. We did not culture samples with *Legionella* spp. infection, so we were unable to perform strain typing to confirm whether the clusters were caused by related strains. The remaining 6 patients from Klerksdorp-Tshepong Hospital Complex appeared to have sporadic infections.

Author affiliations: National Institute for Communicable Diseases, Johannesburg, South Africa (N. Wolter, M. Carrim, C. Cohen, S. Tempia, S. Walaza, P. Sahr, L. de Gouveia, F. Treurnicht, O. Hellferscee, A. von Gottberg); University of the Witwatersrand, Johannesburg (N. Wolter, M. Carrim, C. Cohen, S. Walaza, L. de Gouveia, A. von Gottberg); US Centers for Disease Control and Prevention, Pretoria, South Africa (S. Tempia); University of Pretoria, Pretoria (P. Sahr); US Centers for Disease Control and Prevention, Atlanta, Georgia, USA (A.L. Cohen, A.J. Benitez, J.M. Winchell); Pietermaritzburg Metropolitan Hospitals, Pietermaritzburg, South Africa (H. Dawood); University of KwaZulu-Natal, Pietermaritzburg (H. Dawood); Klerksdorp-Tshepong Hospital Complex, Klerksdorp, South Africa (E. Variava)

DOI: http://dx.doi.org/10.3201/eid2201.150972

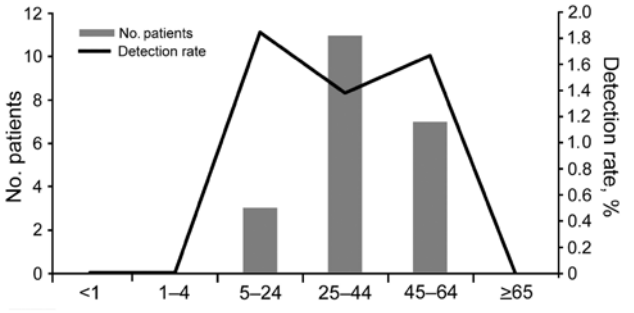

Figure 1. Number of case-patients and detection rate for *Legionella* spp. infections, by age group, South Africa, June 2012–September 2014 (N = 1,803).

Legionella patients resided in different areas or communities within the cities of Pietermaritzburg and Klerksdorp. Epidemiologic investigation revealed exposure to several potential sources of infection, such as waste management, air conditioners, plumbing, mining, and swimming pools; however, no common exposure could be identified, so environmental sampling and testing were not performed.

Fifteen (75%) of 20 *Legionella* patients with known HIV status were infected with HIV, and 9 (43%) of the 21 patients tested positive for TB at the admission during which *Legionella* infection was detected. HIV or TB infection, or both, was detected in 18 (90%) of 20 patients with known HIV and TB status. A history of active TB before the admission during which *Legionella* was detected was reported for 14 (82%) of 17 patients. For 17 *Legionella* spp.–infected patients for whom information was available, additional LD-associated factors included regular alcohol consumption (10 [59%]), cigarette smoking (9 [53%]), asthma (2 [12%]), and heart disease (2 [12%]).

Eighteen (86%) of 21 patients had symptoms >7 days before hospital admission, a delay possibly occurring because many patients were chronically ill (75% were HIV infected and ≥43% had TB). Median duration of hospitalization for *Legionella* patients was 4 days (range 1–35 days), and 1 (9%) patient was admitted to intensive care and survived the illness; 4 (20%) patients died. Antimicrobial drug treatment (in-hospital and discharge medication) was known for 21 patients and included amoxicillin/clavulanic acid (16 [76%]), anti-TB medications (15 [71%]), cotrimoxazole (7 [33%]), cefuroxime/ceftriaxone (5 [24%]), and erythromycin (5 [24%]).

Legionella spp. isolates were identified for 2 patients as *L. pneumophila* serogroup 1 and *L. longbeachae*. Species could not be determined for 19 patients because of low bacterial loads in their specimens. Of the 21 patients with *Legionella*-positive sputum specimens, co-infections were detected in 14 (67%). Co-infecting pathogens were *Mycobacterium tuberculosis* (9 [43%]), rhinovirus (6 [29%]), respiratory syncytial virus (2 [10%]), adenovirus (2 [10%]), *Bordetella pertussis* (1 [5%]), and *Streptococcus pneumoniae* (1 [5%]).

Legionella spp. detection rates in this study were similar to those described in other countries (*4*). However, age distribution tended toward younger adults, not the elderly, the population previously reported as most affected (*4*). Men and women were evenly distributed in our study, although a substantial male predominance is common for LD (*2,4*). Differences in age and gender distributions, compared with distributions in other studies, likely result from high HIV and TB prevalence among younger adults in our study population. LD is typically associated with summer because warm and wet conditions promote bacterial replication (*2,4*). Longer periods of surveillance are needed to establish seasonality of LD in South Africa.

Clinically, patients with LD in this study were likely to be HIV-infected, chronically ill persons with suspected or confirmed TB and were therefore usually treated for TB infection and discharged. HIV-induced immune suppression and lung damage because of biologic or chemical agents likely increased their susceptibility to *Legionella* infections. Cases of LD and TB occurring simultaneously have been previously described (*5–7*). *Legionella* infection in populations with HIV or TB co-infections may cause acute exacerbation of respiratory symptoms, prompting patients to seek hospital care.

In South Africa, treatment for CAP is usually penicillin or ampicillin for adults <65 years of age and amoxicillin/clavunate or cefuroxime for elderly or HIV-infected adults (*8*). However, treatment for LD should include a macrolide or fluoroquinolone (*4*). Only one fourth of *Legionella* patients in this study received appropriate treatment, likely because of clinical inability to distinguish LD from other forms of pneumonia and because clinicians rarely consider *Legionella* when they lack access to diagnostic testing and local prevalence data. This problem is further compounded by the high prevalence of HIV and TB in South Africa. Anti-TB treatment, which was

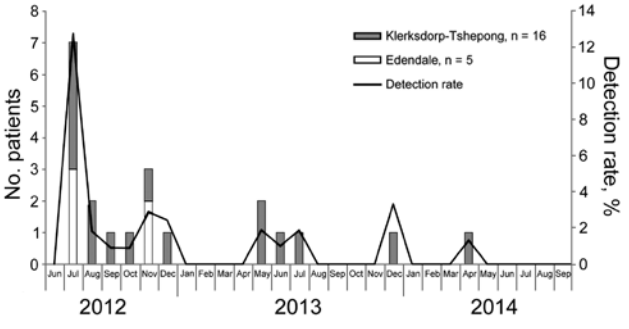

Figure 2. Number of case-patients and detection rate of *Legionella* spp. infections, by month and year, for Edendale Hospital and Klerksdorp-Tshepong Hospital Complex, South Africa, June 2012–September 2014 (N = 1,805).

administered to more than two thirds of the *Legionella* patients, would have had therapeutic benefits; rifampin has been shown to have activity against *Legionella* spp. (*9,10*). However, suboptimal treatment of *Legionella* patients with co-infections likely contributed to a case-fatality ratio (20%) more than twice that for all SRI patients (8%) (*4,11*). Lack of appropriate treatment of patients with CAP in South Africa for atypical pathogens has been described ((*12*).

Conclusions

In South Africa, patients with LD often have chronic illness caused by co-infections such as HIV and TB at time of admission. *Legionella* infections in most patients were undiagnosed, and patients were suboptimally treated for TB or more typical causes of CAP. Increased awareness and improved diagnostic testing could result in earlier diagnosis, appropriate treatment, and improved outcomes for these patients. In addition to routine diagnostics, surveillance for LD should be performed on an ongoing basis for rapid identification and response to outbreaks.

Acknowledgments

We thank Iain Kennedy, who assisted with the epidemiologic investigation, and Makatisane Papo for his help with data management. We also thank all members involved in the Severe Acute Respiratory Illness study for the collection of specimens and data management.

This work was supported by the National Health Laboratory Service, South Africa, and the US Centers for Disease Control and Prevention (CDC-RFA-GH12-00403). A. von G. received grants from the US Centers for Disease Control and Prevention and Pfizer-South Africa. H.D. received honoraria or travel grants from Merck Sharp & Dohme–South Africa; Novartis–South Africa; and Pfizer–South Africa.

Dr. Wolter is a senior medical scientist in the Centre for Respiratory Diseases and Meningitis at the National Institute for Communicable Diseases in Johannesburg and is a specialist in molecular microbiology. Her research interests include the diagnosis and epidemiology of respiratory pathogens.

References

1. Dlamini SK, Mendelson M. Atypical pneumonia in adults in southern Africa. South Afr J Epidemiol Infect. 2012;27:5–9.
2. Marston BJ, Lipman HB, Breiman RF. Surveillance for Legionnaires' disease. Risk factors for morbidity and mortality. Arch Intern Med. 1994;154:2417–22. http://dx.doi.org/10.1001/archinte.1994.00420210049006
3. Thurman KA, Warner AK, Cowart KC, Benitez AJ, Winchell JM. Detection of *Mycoplasma pneumoniae, Chlamydia pneumoniae*, and *Legionella* spp. in clinical specimens using a single-tube multiplex real-time PCR assay. Diagn Microbiol Infect Dis. 2011; 70:1–9. http://dx.doi.org/10.1016/j.diagmicrobio.2010.11.014
4. Phin N, Parry-Ford F, Harrison T, Stagg HR, Zhang N, Kumar K, et al. Epidemiology and clinical management of Legionnaires' disease. Lancet Infect Dis. 2014;14:1011–21. http://dx.doi.org/10.1016/S1473-3099(14)70713-3
5. Hamedani P, Hafeez S, Ali J, Memon R, Ali S, Ali M, et al. Acute and chronic Legionnaires' disease and co-existent tuberculosis: a trial of erythromycin. Curr Med Res Opin. 1989; 11:510–7. http://dx.doi.org/10.1185/03007998909110462
6. Milder JE, Rough RR. Concurrent Legionnaires' disease and active pulmonary tuberculosis. Am Rev Respir Dis. 1982;125:759–61.
7. Hamedani P, Hafeez S, Ali J, Memon R, Ali S, Ali M, et al. Acute and chronic Legionnaires' disease and co-existent tuberculosis: a trial of erythromycin. Curr Med Res Opin. 1989;11:510–7. http://dx.doi.org/10.1185/03007998909110462
8. Working Group of the South African Thoracic Society. Management of community-acquired pneumonia in adults. S Afr Med J. 2007;97:1296–306.
9. Klein NC, Cunha BA. Treatment of legionnaires' disease. Semin Respir Infect. 1998;13:140–6.
10. Vesely JJ, Pien FD, Pien BC. Rifampin, a useful drug for nonmycobacterial infections. Pharmacotherapy. 1998;18:345–57.
11. Heath CH, Grove DI, Looke DF. Delay in appropriate therapy of *Legionella pneumonia* associated with increased mortality. Eur J Clin Microbiol Infect Dis. 1996;15:286–90. http://dx.doi.org/10.1007/BF01695659
12. Nyamande K, Lalloo UG. Poor adherence to South African guidelines for the management of community-acquired pneumonia. S Afr Med J. 2007;97:601–3. PubMed

Address for correspondence: Nicole Wolter, Centre for Respiratory Diseases and Meningitis, National Institute for Communicable Diseases, Private Bag X4, Sandringham, 2131, Gauteng, South Africa; email: nicolew@nicd.ac.za

Article DOI: http://dx.doi.org/10.3201/eid2201.150972

Legionnaires' Disease in South Africa, 2012–2014

Technical Appendix

Materials and Methods

Study Design

We conducted a prospective, hospital-based, observational study as part of Severe Respiratory Illness (SRI) surveillance from June 2012–September 2014 at 2 sites in South Africa: Klerksdorp-Tshepong Hospital Complex, Klerksdorp, North West Province; and Edendale Hospital, Pietermaritzburg, KwaZulu-Natal Province.

A case of SRI was defined as a person hospitalized with symptoms of any duration and meeting age-specific clinical inclusion criteria as follows: children from 2 days to <3 months of age with physician-diagnosed sepsis or lower respiratory tract infection (LRTI); children from 3 months to <5 years of age with physician-diagnosed LRTI; and patients ≥5 years of age who meet a modified World Health Organization case definition for SRI (*1*): 1) fever (>38°C) or reported fever; 2) cough or sore throat; and 3) shortness of breath or difficulty breathing, with or without clinical or radiographic findings of pneumonia. In addition, we enrolled patients with a clinical diagnosis of suspected tuberculosis (TB). Chest x-rays are not performed routinely on LRTI patients at the site hospitals, so radiologic confirmation of pneumonia could not be obtained. A case of Legionnaires' disease was defined as a person hospitalized with SRI and positive results for *Legionella* spp. infection determined by PCR from an induced sputum or nasopharyngeal (NP) specimen. A standardized questionnaire was used to collect demographic and clinical information, such as age, sex, regular alcohol or cigarette use, mining exposure, underlying illness, prior antimicrobial drug use, duration of symptoms, length of hospitalization, in-hospital antimicrobial drug treatment, treatment with supplemental oxygen, admission to an intensive care unit, and outcome.

Specimen Collection

NP, induced sputum, and whole blood samples were collected. NP specimens, including combined NP and oropharyngeal swabs (FLOQSwabs, Copan Diagnostics, Murrieta, CA, USA) for patients ≥5 years of age or NP aspirates for children <5 years of age were placed in universal transport medium (Copan Diagnostics) and transported at 4°C to the National Institute for Communicable Diseases, Johannesburg, South Africa. As of July 2013, sputum samples were stored at −20°C and transported on dry ice.

Detection of *Legionella* spp.

Sputum samples were digested with dithiothreitol (Roche Diagnostics, Mannheim, Germany). Total nucleic acids were extracted from 200 µL of sample by using the MagNA Pure 96 instrument (Roche) and MagNA Pure 96 DNA and Viral NA SV kit (Roche). Nucleic acids were eluted into 100 µL of elution buffer and stored at −20°C.

NP and induced sputum samples were tested for *Legionella* spp. by using a multiplex real-time PCR assay that also detected *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, and the human *RNaseP* gene (*2*). Twenty-five µL reactions were performed by using PerfeCTa Multiplex qPCR Supermix (Quanta Biosciences, Gaithersburg, MD, USA), 6.5 µL DNA, and primers and probes, as previously described (*2*). Samples with a C_t-value >45 were recorded as negative. PCR was performed by using an Applied Biosystems 7500 Fast real-time PCR instrument (Thermo-Fisher Scientific, Waltham, MA).

Legionella spp. Identification

Legionella-positive specimens were further tested in 2 real-time PCR assays. The first assay confirmed *Legionella* spp. and identified *L. pneumophila* and *L. pneumophila* serogroup 1 (*3*). The second assay identified *L. longbeachae*. The reaction mix comprised of 12.5 µL of PerfeCTa Multiplex qPCR Supermix (Quanta Biosciences), 5 µL DNA, 25 µmol/L primers (LLB-F –TGGTTTTCGAAATCATCAGTATGC, LLB-R-CTGTCTAAAACACTTCTCTCCCGATA) and 5 µM probe (Quasar670-TTTAATTTAGTTCCCACCAGCAAGGATGGC-BHQ3) made to a final volume of 25 µL. Cycling conditions were as follows: 1 cycle of 95°C for 10 min, followed by 50 cycles of 95°C for 15 s and 60°C for 1 min.

TB Testing

TB testing was conducted at the site laboratory; additional testing was done at the NICD. Sputum samples were tested for *Mycobacterium tuberculosis* by smear microscopy,

culture or PCR. Tests were performed as follows: smear microscopy by using fluorescence auramine staining for acid-fast bacilli; liquid media by using BD Bactec MGIT 960 (Beckton, Dickinson, Franklin Lakes, NJ, USA) for culture; and TB PCR by using the Xpert MTB/RIF system (Cepheid, Sunnyvale, CA, USA). Positive cultures were identified as *M. tuberculosis* complex by using Ziehl-Neelsen staining and antigen testing.

Detection of Other Respiratory Pathogens

SRI patients were tested for additional respiratory pathogens. NP specimens were tested for specific respiratory viruses (i.e., influenza, adenovirus, enterovirus, rhinovirus, human metapneumovirus, respiratory syncytial virus, and parainfluenza types 1–3) by multiplex real-time PCR (*4*). NP and induced sputum specimens were tested for *Bordetella pertussis* (*5*), and blood specimens were tested for *Streptococcus pneumoniae* (*6*) and *Haemophilus influenzae* (*7*).

Determination of HIV Status

HIV results were obtained from a combination of 2 sources: patient clinical records when available and for consenting patients, a blind, linked dried-blood spot tested at NICD. For patients for whom both results were available, the NICD result was used. Testing included HIV ELISA testing for patients ≥18 months of age and PCR testing for children <18 months of age.

Epidemiologic Case Investigation

An epidemiologic investigation was conducted retrospectively for *Legionella*-positive patients who could be traced and included collection of additional information from the patient or a close relative for a deceased patient. Additional information included potential *Legionella* exposure, past medical history, factors associated with disease, and discharge medication.

Statistical Analysis

As sputum specimens are the optimal specimen type for *Legionella* spp. detection, analyses were performed for patients who had a sputum sample collected. Detection rate was defined as the number of *Legionella* case-patients among all SRI case-patients with sputum samples collected and tested.

Ethical Approval

The protocol was approved by the Universities of the Witwatersrand (M081042) and KwaZulu-Natal (BF157/08). The U.S. Centers for Disease Control and Prevention deemed the study a non-research, surveillance activity.

References

1. World Health Organization. WHO interim global epidemiological surveillance standards for influenza. Geneva: The Organization; 2012 [cited 2013 Feb 14]. http://www.who.int/influenza/resources/documents/INFSURVMANUAL.pdf

2. Thurman KA, Warner AK, Cowart KC, Benitez AJ, Winchell JM. Detection of *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, and *Legionella spp*. in clinical specimens using a single-tube multiplex real-time PCR assay. Diagn Microbiol Infect Dis. 2011;70:1–9. http://dx.doi.org/10.1016/j.diagmicrobio.2010.11.014

3. Benitez AJ, Winchell JM. Clinical application of a multiplex real-time PCR assay for simultaneous detection of *Legionella* species, *Legionella pneumophila*, and *Legionella pneumophila* serogroup 1. J Clin Microbiol. 2013;51:348–51. http://dx.doi.org/10.1128/JCM.02510-12

4. Pretorius MA, Madhi SA, Cohen C, Naidoo D, Groome M, Moyes J, et al. Respiratory viral coinfections identified by a 10-plex real-time reverse-transcription polymerase chain reaction assay in patients hospitalized with severe acute respiratory illness–South Africa, 2009–2010. J Infect Dis. 2012;206(Suppl 1):S159–65. http://dx.doi.org/10.1093/infdis/jis538

5. Tatti KM, Sparks KN, Boney KO, Tondella ML. Novel multitarget real-time PCR assay for rapid detection of *Bordetella* species in clinical specimens. J Clin Microbiol. 2011;49:4059–66. http://dx.doi.org/10.1128/JCM.00601-11

6. Carvalho M, Tondella ML, McCaustland K, Weidlich L, McGee L, Mayer LW, et al. Evaluation and improvement of real-time PCR assays targeting *lytA*, *ply*, and *psaA* genes for detection of pneumococcal DNA. J Clin Microbiol. 2007;45:2460–6. http://dx.doi.org/10.1128/JCM.02498-06

7. Maaroufi Y, De Bruyne JM, Heymans C, Crokaert F. Real-time PCR for determining capsular serotypes of *Haemophilus influenzae*. J Clin Microbiol. 2007;45:2305–8. http://dx.doi.org/10.1128/JCM.00102-07

Technical Appendix Table. Demographic, epidemiologic, and clinical characteristics of patients positive for *Legionella*, South Africa, June 2012–September 2014 (N = 22)*

Case no.	Hospital, admission date	Sex, age, y	Symptom duration, d†	Potential exposure to contaminated water source	HIV	TB‡	Risk factors§	Hospital duration, d (treatment¶)	Outcome
E1#	Edendale, 28 Jun 2012	M, 56	4	Worked/lived at waste management compound, compost exposure	Unk	Sput unav	Heart disease, alcohol	35 (TB, amoxicillin-clavunate)	Survived
E2**	Edendale, 3 Jul 2012	M, 52	Unk	Unk	Unk	Sput –; treated for TB in past year	None	6 (Amoxicillin-clavunate)	Died
E3**	Edendale, 9 Jul 2012	F, 55	19	Unk	+	Sput +	None	3 (Amoxicillin-clavunate)	Died
E4	Edendale,16 Jul 2012	M, 20	13	Attended school	–	Sput –	Former smoker, alcohol	22 (TB, amoxicillin-clavunate)	Survived
E5**	Edendale, 6 Nov 2012	F, 41	66	Unk	+	Sput –	None	6 (Cefuroxime)	Survived
E6	Edendale, 13 Nov 2012	F, 32	8	Cleaner at hospital/air conditioner maintenance	+	Sput +	None	7 (TB, amoxicillin-clavunate, cefuroxime)	Survived
T1**	Tshepong, 3 Jul 2012	F, 34	30	Unk	+	Sput +	None	1 (HIV,TB, amoxicillin-clavunate, cotrimoxazole)	Died out of hospital
T2**	Tshepong, 4 Jul 2012	F, 46	30	Unk	+	Sput –	None	5 (TB)	Survived
T3	Tshepong, 10 Jul 2012	F, 32	61	None known	+	Sput +	Heart disease	2 (HIV, TB, ceftriaxone, cefuroxime)	Survived
T4	Tshepong, 30 Jul 2012	M, 44	1	Gardening/ plumbing work, compost exposure	–	Sput –	Current smoker, alcohol	2 (TB, erythromycin)	Survived
T5	Tshepong, 14 Aug 2012	M, 34	10	Mining, gardening, renovation work	+	Sput +	Asthma, former smoker, alcohol	1 (HIV, erythromycin, amoxicillin-clavunate)	Survived
T6	Tshepong, 16 Aug 2012	F, 19	13	None known	–	Sput +	Alcohol	3 (TB, amoxicillin-clavunate)	Survived
T7	Tshepong, 20 Sep 2012	F, 50	Unk	None known	+	Sput –	Asthma, current smoker (snuff), alcohol	5 (HIV, TB, cotrimoxazole, erythromycin, amoxicillin-clavunate)	Survived
T8	Tshepong, 21 Oct 2012	M, 25	14	Sewage exposure	–	Sput +	Alcohol	1 (TB, amoxicillin-clavunate, cefuroxime)	Survived
T9	Tshepong, 5 Nov 2012	F, 39	188	None known	+	Sput –	Current smoker (snuff)	11 (HIV, TB, cotrimoxazole)	Survived
T10	Tshepong, 6 Dec 2012	M, 55	21	None known	+	Sput –	Former smoker	1 (TB, amoxicillin-clavunate, cefuroxime, cotrimoxazole, erythromycin)	Survived
T11	Tshepong, 6 May 2013	F, 52	22	Hospital admission 22 Apr 2013–1 May 2013	+	Sput –	None	3 (HIV, cotrimoxazole, amoxicillin-clavunate)	Died
T12	Tshepong, 9 May 2013	M, 59	13	Visited wife in hospital Apr 2013	+	Sput –	Current smoker, alcohol	5 (Amoxicillin-clavunate)	Survived
T13	Tshepong, 11 Jun 2013	M, 42	10	Building and cleaning of dams/ swimming pools	+	Sput +	Former smoker, alcohol	Unk (HIV, TB, amoxicillin-clavunate,	Survived

Case no.	Hospital, admission date	Sex, age, y	Symptom duration, d†	Potential exposure to contaminated water source	HIV	TB‡	Risk factors§	Hospital duration, d (treatment¶)	Outcome
								cotrimoxazole)	
T14	Tshepong, 1 Jul 2013	M, 22	2	Home renovation Jan–Mar 2013	–	Sput +	Former smoker	2 (TB, amoxicillin-clavunate)	Survived
T15	Tshepong, 5 Dec 2013	M,38	3	None known	+	Sput –	Current smoker, alcohol	4 (TB, cotrimoxazole, amoxicillin-clavunate)	Survived
T16	Tshepong, 16 Apr 2014	M, 38	13	Mining exposure	+	Sput –	Former smoker	27 (HIV, erythromycin)	Survived

*None known, patient or family member did not report exposure to any water source listed on questionnaire; TB, *Mycobacterium tuberculosis* infection; Unk, unknown: information could not be obtained because interviews could not be conducted; +, positive; –, negative; unav, unavailable; Sput, sputum.
†Symptom duration includes time from symptom onset until hospital admission
‡TB status was determined by sputum test with positive (Sput +) or negative (Sput –) results. Sputum test was unavailable for patient E1.
§Risk factors assessed were asthma, lung disease, kidney disease, heart disease, liver disease, diabetes, cancer, emphysema, former/current cigarette smoking, and current alcohol consumption.
¶Includes in-hospital and discharge treatment, mostly for HIV or TB infection or both.
#*Legionella* spp. identified with nasopharyngeal specimen.
**Retrospective epidemiologic investigation not possible because patient died or patient or family member could not be traced.